VGEN.P-056-US PATENT APPLICATION

What is Claimed is:

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1. A method for assignment of base numbers to peaks within an experimental DNA sequencing data trace derived from the separation of experimental DNA sequencing fragments, comprising the steps of:

- (a) obtaining one or more reference DNA sequencing data traces derived from the separation of reference DNA sequencing fragments reflecting the position of at least one base in a reference polynucleotide of known sequence;
- (b) evaluating the reference DNA sequencing data traces to determine a corrected time scale indicative of migration times at which peaks should occur;
- (c) sampling the experimental DNA sequencing data trace at time points determined by the corrected time scale, and
- (d) assigning a base number to each peak found in the experimental DNA sequencing data trace based upon the corrected time scale.
- 2. The method of claim 1, wherein the step of evaluating the reference DNA sequence data traces includes the steps of:
- (i) identifying a plurality of peaks in the reference DNA sequencing data traces, and creating a data table containing the number of each peak based on the known sequence of the polynucleotide, and the position of each peak in the reference DNA sequencing data trace;
- (ii) identifying a set of coefficients for a polynomial effective to substantially linearize a plot of peak number versus separation between adjacent peaks; and
- (iii) creating from the coefficients and the polynomial a corrected time scale which reflects the positions at which a peak should occur at any given point in a sequencing data trace.

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1	Suby	3.	The method of claim 1, wherein the experimental DNA sequencing data
2	trace and a fir	rst refer	ence DNA sequencing data trace are derived from analysis of sequencing
3	fragments in	a comm	on lane of a sequencing gel.
1		4.	The method of claim 1, wherein a plurality of reference DNA sequencing
2	data traces are	e obtain	ed, each derived from the separation of the same set of reference DNA
3	sequencing fr	agment	S.
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1	AS polynomial.	5.	The method of claim 1, wherein the polynomial is a third or higher order
2	A5 polynomial.		
	•		
1		6.	The method of claim 1, wherein a defined number of bands are selected
2	for evaluation	n from e	each of the reference DNA sequencing data traces.
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		7.	The method of claim 6, wherein the defined number of bands selected is
2	from 3 to 40		
Į.			
+ 2 4 7 7		8.	The method of claim 6, wherein the defined number of bands is at least
=	equal to the o		the polynomial, plus 1.
#	equal to the e	nuci oi	the polynomia, pras 1.
1		9.	The method of claim 1, wherein base numbers are assigned to peaks
2	within a nlur		experimental DNA sequencing data traces derived from the separation of
	-	_	
3		DINA S	requencing fragments indicative of the positions of a plurality of types of
4	bases.		

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1		10.	The method of claim 9, wherein base numbers are assigned to peaks
2	within four experimental DNA sequencing data traces derived from the separation of		
3	experimental	DNA s	equencing fragments indicative of the positions of four types of bases.
1		11.	A method for evaluating the sequence of a target polynucleotide,
2	comprising th	ne steps	of:
3		(a)	obtaining one or more experimental DNA sequencing data traces derived
4	from the sepa	ration c	of experimental DNA sequencing fragments reflecting the position of at least
5	one base in th	ne target	polynucleotide and one or more reference DNA sequencing data traces
6	derived from	the sepa	aration of reference DNA sequencing fragments reflecting the position of at
6	least one base	e in a re	ference polynucleotide of known sequence;
8		(b)	evaluating the reference DNA sequencing data traces to determine a
9	corrected time scale indicative of migration times at which peaks should occur;		
<u></u>		(c)	sampling the experimental DNA sequencing data traces at time points
ŧ	determined b	y the co	rrected time scale, and
2		(d)	assigning a base number to each peak found in the experimental DNA
3	sequencing d	ata trace	es based upon the corrected time scale, thereby obtaining information about
	the sequence	of the ta	arget polynucleotide.
1		12.	The method of claim 11, wherein the step of evaluating the reference DNA
2	sequence data	a traces	includes the steps of:
3		(i)	identifying a plurality of peaks in the reference DNA sequencing data
4	traces, and cr	eating a	data table containing the number of each peak based on the known
5	sequence of t	he poly	nucleotide, and the position of each peak in the reference DNA sequencing
6	data trace;		
7		(ii)	identifying a set of coefficients for a polynomial effective to substantially

linearize a plot of peak number versus separation between adjacent peaks; and

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9		(iii)	creating from the coefficients and the polynomial a corrected time scale
10	which reflects	s the po	sitions at which a peak should occur at any given point in a sequencing data
11	trace.		
1	l	13.	The method of claim 11, wherein the reference DNA sequencing traces
2 cut	and the exper	imental	DNA sequencing data trace are derived from analysis of sequencing
3 Ale	fragments in a	a comm	DNA sequencing data trace are derived from analysis of sequencing on sequencing gel.
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1		14.	The method of claim 13, wherein the experimental DNA sequencing data
2	trace and a fir	st refere	ence DNA sequencing data trace are derived from analysis of sequencing
			on lane of the common sequencing gel.
l		15.	The method of claim 11, wherein a plurality of reference DNA sequencing
2	data traces are	obtain	ed, each derived from the separation of the same set of reference DNA
3	sequencing fra	agments	3.
รูป 2 คา	polynomial.	16.	The method of claim 11, wherein the polynomial is a third or higher order
1		17.	The method of claim 11, wherein a defined number of bands are selected
2	for evaluation	from ea	ach of the reference DNA sequencing data traces.
1		18.	The method of claim 17, wherein the defined number of bands selected is
2	from 3 to 40.		
1		19.	The method of claim 17, wherein the defined number of bands is at least
2	equal to the or	der of the	he polynomial, plus 1.

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1	20. The method of claim 11, wherein base numbers are assigned to peaks		
2	within a plurality of experimental DNA sequencing data traces derived from the separation of		
3	experimental DNA sequencing fragments indicative of the positions of a plurality of types of		
4	bases.		
1	21. An apparatus for evaluating the sequence of a target polynucleotide,		
2	comprising:		
3	(a) an input for receiving information about one or more experimental DNA		
4	sequencing data traces derived from the separation of experimental DNA sequencing fragments		
5	reflecting the position of at least one base in the target polynucleotide and one or more reference		
6	DNA sequencing data traces derived from the separation of reference DNA sequencing		
ī	fragments reflecting the position of at least one base in a reference polynucleotide of known		
	sequence;		
9	(b) a processor, operatively programmed to evaluate the reference DNA		
10	sequencing data traces to determine a corrected time scale indicative of migration times at which		
14	peaks should occur;		
	(c) a processor, operatively programed to sample the experimental DNA		
1.3	sequencing data traces at time points determined by the corrected time scale;		
14	(d) a processor, operatively programmed to assign a base number to each peak		
15	found in the experimental DNA sequencing data traces based upon the corrected time scale,		
16	thereby obtaining information about the sequence of the target polynucleotide; and		
17	(e) an output for communicating the information about the sequence of the		
18	target polynucleotide.		
1	22. The apparatus of claim 21, wherein the processor programmed to evaluate		
2	the reference DNA sequence data traces is programmed to perform the steps of:		

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(1) identifying a plurality of peaks in the reference DNA sequencing data
traces, and creating a data table containing the number of each peak based on the known
sequence of the polynucleotide, and the position of each peak in the reference DNA sequencing
data trace;
(ii) identifying a set of coefficients for a polynomial effective to substantially
linearize a plot of peak number versus separation between adjacent peaks; and
(iii) creating from the coefficients and the polynomial a corrected time scale
which reflects the positions at which a peak should occur at any given point in a sequencing data
trace.